

## Drycore Appears to Result from an Interaction between *Rhizoctonia solani* and Wireworm (*Agriotes* ssp.)—Evidence from a 3-Year Field Survey

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**Abstract** Drycore is an important quality deficiency in Europe especially in organic potato production and after grass clover leys. The drycore symptom is attributed to *Rhizoctonia solani* Kühn (teleomorph: *Thanatephorus cucumeris* (Frank) Donk). In the framework of a 3-year survey (2001–2003), data concerning potato quality, crop rotation, management, and site parameters were analysed from 185 potato fields. The hypothesis was tested that injuries on potatoes caused by wireworms facilitate the penetration of *R. solani* and favour the formation of drycore. Analysis of variance showed a significant influence of wireworm damage, seed quality, and grass clover leys in the crop on the level of drycore damage. On fields which had both a low occurrence of black scurf on the seed tubers and a low occurrence of wireworm damage at harvest, significant drycore damage was never observed. The relative risk for drycore damage on tubers was significantly higher if black scurf or wireworm damage was on the same tuber. In contrast, no higher risk for drycore was observed on tubers with slug damage. Abiotic factors like farm manure application, organic matter content, texture, and pH of the soil also had no significant influence on the level of drycore. Thus, the wounding of potatoes by wireworm could be confirmed as the major variable for drycore. The mode of action has to be clarified under controlled conditions.

**Keywords** Drycore · *Rhizoctonia solani* · Slugs · *Solanum tuberosum* · Wireworm

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## Introduction

Drycore is one of the main reasons that potato lots do not meet the demanded quality standard in organic potato production but is also the cause for important losses in integrated and conventional farming systems (Keiser et al., unpublished results). The restricted brown cavities up to several millimetre deep with a diameter of 3 to 6 mm are attributed to *Rhizoctonia solani* Kühn (teleomorph: *Thanatephorus cucumeris* (Frank) Donk) (Ramsey 1917; Müller 1925; Grütte 1940; Schwinn 1961). *R. solani* is a soilborne plant pathogen affecting many agricultural crops worldwide (Ogoshi 1987; Sneh et al. 1991). On potatoes, the disease causes delayed emergence, lesions on stems (stem canker) and stolons and sclerotial formation on tubers (black scurf) (Baker 1970; Anderson 1982). Attacks on stolons may induce the development of misshapen tubers and tubers with growth cracks (Weinhold et al. 1978; Jeger et al. 1996). Differences in susceptibility to *R. solani* can be seen between cultivars (Scholte 1989), but no resistant cultivars have been identified so far. Buhr (1989) showed that reactions to tuber deformations varied significantly and reproducibly between cultivars. For all other symptoms, the influence of environmental conditions was bigger than the influence of cultivars.

To date, 14 anastomosis groups (AG 1 to AG 13 and AG BI) have been identified and seven of them (AG 1, AG 2, AG 3, AG 4, AG 6, AG 8 and AG 9) are divided into subgroups that differ in pathology, biochemical or genetic characteristics (Anderson 1982; Cubeta and Vilgalys 1997; Gonzalez et al. 2001; Carling et al. 2002). AG 2-1, 3, 4, 5, 8 and 9 are capable of causing moderate to extensive damage to potato plants. AG 3 has been known as the most host-specific type of *R. solani* causing a disease of potato. It is by far the most important cause of black scurf because most of the sclerotia isolated from potato tubers belong to AG 3 (Carling and Leiner 1986; Chand and Logan 1983; Campion et al. 2003).

In a survey encompassing different farming systems in Switzerland, organic fields showed considerably more serious drycore damage than integrated fields. Rainfall and soil type had no impact on this damage (Keiser et al., unpublished results). This contradicts the postulate of Schwinn (1961) that the formation of drycore symptoms is favoured under wet soil conditions. Although some relationship between drycore damage and black scurf existed, tubers from plants with heavy seed infections and *Rhizoctonia* symptoms on the shoots could be absolutely free of drycore. However, drycore symptoms on tubers were never observed on plants without any symptoms of *R. solani*. Thus, no strong correlation was observed between black scurf level on harvested tubers and the appearance of drycore on the same tuber. Therefore, the infection of a plant with *R. solani* cannot be the only factor necessary for the formation of drycore. Intriguingly, wireworms and their damage on tubers seemed to be directly or indirectly linked to drycore occurrence (Keiser et al., unpublished results). Wireworms are the larvae of click beetles (*Elateridae*). The most important pest species are those belonging to the genus *Agriotes*. Three species, *Agriotes lineatus* (L.), *Agriotes obscurus* (L.) and *Agriotes sputator* (L.) are responsible for the vast majority of wireworm attacks in Britain (Parker and Howard 2001). In Switzerland, *A. obscurus* (L.) and *A. lineatus* (L.) are the most important species in the north of the Alps, whereas *Agriotes littigiosus* and *A. obscurus* are dominant in the south of the Alps (Jossi and Bigler 1997). Crops susceptible to injury are cereals,

maize, potatoes, sugar beet and vegetables (Glogoza 2001). In potatoes, progeny tubers can be severely damaged. Infested tubers have narrow tunnels resulting in a lower market value or even total refusal of the potato lot. Wireworm damage may be confused with slug damage. However, slug often hollow out large cavities within the tuber flesh, whereas wireworms do not (Gratwick 1989). In this study, the hypothesis was tested that injuries caused by wireworm or other means may facilitate the penetration of *R. solani* into the tuber and favour the formation of drycore symptoms. Data on crop rotation, management and site parameters of potato fields from a large survey were analysed to check this hypothesis.

## Material and Methods

### Description of the Experimental Sites and Experimental Design

In 2002 and 2003, potato fields from different farming systems were evaluated across the major potato-growing regions of Switzerland. In the survey, 185 plots were included (40 organic, 42 integrated and 103 conventional). All relevant data concerning crop rotation, management and site parameters were recorded and around 20 external quality deficiencies on the harvested tubers were assessed. Potato growers filled out a questionnaire with all relevant data concerning crop rotation and management for all fields included in the project. Soil samples of all fields in the project were analysed according to the reference method of the Swiss federal research stations Agroscope (Düngeberatung 2004): clay, silt and sand content (finger test), soil type, pH (H<sub>2</sub>O), organic matter (analytic) and ammonium acetate EDTA extractable P, K and Mg content.

Soil types were mostly sandy loam (61%) and clay loam (22%). Soil pH varied widely between 5.4 and 8.2. Organic matter contents were between 1.7% and 6.8% in 80% of the fields; only 5% had more than 10%. The soil nutrient status ranged between “adequate” and “enriched” for about 75% of the P, K and Mg analytical results and was “slightly deficient” in about 24% of the cases.

The long-term annual mean temperature (1961 until 1990) in the main potato-growing areas of Switzerland was between 8.0 and 9.8 °C. Year 2002 and especially 2003 belonged to the hottest years ever with mean temperatures of 1 to 2.5 °C higher than the long-term mean. The annual precipitation varied strongly among years and regions. The annual precipitation was 125% (1,000–1,300 mm) of the long-term annual mean in 2002. In contrast, 2003 was extraordinary dry and hot in all regions with 600 to 750 mm (75% of the long-term mean). In 2002 and 2003, all fields were planted before the end of April (Bundesamt für Meteorologie und Klimatologie 2006).

Four main varieties from the Swiss variety list were selected for the project. Charlotte (maturity: early) and Agria (early to intermediate) are the most important varieties in all three farming systems. Charlotte is the most popular variety for the fresh market, whereas Agria is produced for both the fresh market and the processing industry (French fries). Bintje (early to intermediate) is still important for the fresh market in the integrated and conventional farming systems. Eba (intermediate to late) is used for French fry production in the processing industry.

Intervals between two potato crops were at least 3 years for 25% of the fields and five or more years for the other 75%. In the organic farming system, the most frequent preceding crops were grass clover ley (43%), followed by cereals (24%) and vegetables (22%). On integrated and conventional fields, grass clover leys were much less frequent (23% and 10%, respectively), most frequent were cereals (51 and 46%) followed by maize and sugar beet.

The average number of fungicide treatments against late blight (*Phytophthora infestans*) was 7.6 for conventional and 7.4 for integrated fields. On organic fields, the average number of treatments was 3.6, mainly with copper. In the integrated system, potato seed tubers were treated with a fungicide against *R. solani* on 36% of the fields, compared to 59% in the conventional system. Seventy-five percent of all treatments were done at planting, 25% during storage of the seed potatoes. The most frequent product used was Monceren with the active substance penicuron. No treatments with synthetic fungicides are allowed in organic agriculture. Thirty percent of the conventional and integrated fields were treated once and around 10% twice with molluscicide pellets against slugs. In the integrated and conventional system, an insecticide seed treatment was used at least once in the five preceding years to potatoes on 19% and 21%, respectively, of the fields cropped with cereals, sugar beet or corn. Insecticide treatments of seed potatoes or soil applications are not allowed in Switzerland.

Manure was applied on 36% (organic and integrated) and 28% (conventional) of the fields. An application of liquid manure was much more frequent in the organic system (34%) than in the integrated and conventional systems with 13% and 17%, respectively. On 14% (organic), 3% (integrated) and 5% (conventional) of the fields, both solid and liquid manure was applied. Manure was most frequently applied in spring before ploughing. On 48% (integrated) and 50% (conventional) of the fields, no manure was applied compared to 16% in the organic system. On organic fields, over 90% of the total amount of phosphorus, potassium and magnesium per hectare and 70% of nitrogen was applied with manure. In the integrated and conventional farming system, about 50% of phosphorus, 66% of potassium and magnesium and 80% of nitrogen fertilisation were applied with mineral fertilisers.

### Potato Quality Assessment

In 2002 and 2003, 100 tubers were randomly sampled from the seed tuber lots of each field. Tubers were washed and checked for black scurf (*R. solani* Kühn) according to the guidelines of the organisation of the Swiss seed multipliers (Swissem) (SSPV 2002). Just before harvest, samples consisting of 810 tubers were collected on each field. On every fourth ridge of each 0.8 ha field, a total of 270 plants was harvested. Three tubers in the demanded size were randomly sampled from each plant. All tubers were washed before the assessment of quality deficiencies. The number of lesions per tuber caused by wireworm, drycore and slugs was counted. To discriminate between the different lesions, all tubers were peeled with a special knife to a depth of 4 mm, the maximal tolerated depth (Schweizerische Kartoffelkommission 1989). Wireworms cause narrow tunnels up to more than 1 cm deep, whereas slugs often hollow out large cavities within the tuber flesh

(Gratwick 1989). Drycore symptoms are brown cavities up to several millimetre deep with a diameter of 3 to 6 mm. Black scurf on seed tubers and harvested tubers was assessed using grades from 1 (no black scurf) to 7 (75% to 100% of the tuber surface with black scurf).

### Relative Risk for Drycore

To test the within field distribution and possible relations between an influencing factor and drycore, the relative risk for drycore damage on a tuber was calculated for the influencing factors black scurf, wireworm damage and slug damage for 40 fields (totally 32,400 tubers) with a drycore level of more than 5% damaged tubers. This level exceeds the maximal tolerance and leads to price reduction or even rejection of the potato lot. The relative risk is the ratio of the event rate between comparative groups and is calculated with the following equation:  $\text{relative risk} = [(a/a+b)/(c/c+d)]$ , where  $a$  = tubers with drycore and influencing factor is present,  $b$  = tubers without drycore, but influencing factor is present,  $c$  = tubers with drycore but influencing factor is not present and  $d$  = tubers without drycore and no influencing factor. A relative risk  $>1$  means drycore symptoms are more likely to occur if the influencing factor is present on the same tuber.

### Statistical Analysis

To determine the influence of various factors on different quality parameters, the project fields were grouped according to management practice (e.g. farming system and crop rotation) and site parameters (e.g. clay content and pH). Measurement results were statistically checked for normal distribution and comparable variability. Depending on the results, averages or medians were compared with the following methods:

- Two- and higher way analyses of variance. In certain cases,  $\arcsin \sqrt{p}$  transformation was done before analyses of variance. From the full model, all interactions with a probability level  $>0.15$  were removed stepwise and residuals of the final model were tested for normal distribution. Main factors from the final model were removed to test stability of the result.
- $T$  test for two independent samples of normally distributed but heterogeneous data.
- Wilcoxon rank-sum test for not normally distributed data.

In order to have a global  $\alpha$ -level of 0.05 for the family of the tests performed, the Tukey–Kramer test and the Bonferroni–Holm procedure were applied, respectively (Holm 1979). All statistical analysis was done with Number Cruncher Statistical Software 2004 (Kaysville, Utah 84037, UT, USA).

## Results

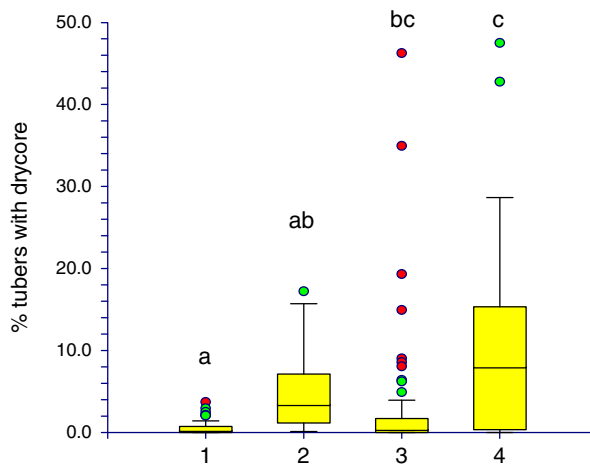
On fields with both a low occurrence of black scurf on the seed tubers and a low occurrence of wireworm damage, important drycore damage of over 5% was never

observed. The significantly highest damage was observed when more than 10% of the planted seed tubers had black scurf and more than 5% of the harvested tubers showed wireworm holes (Fig. 1). Even on fields with heavy black scurf infestation, drycore occurred very rarely without wireworm injuries on the tubers. The analysis of variance for drycore showed a significant influence of wireworm damage, seed quality and grass clover ley as preceding crop on the level of drycore damage (Table 1). Drycore damage was significantly highest on fields with grass clover ley in the 2 years preceding potatoes when more than 20% of seeds were infested with black scurf. Where clean seed was planted on fields without grass clover ley in the two preceding years little drycore damage occurred. No influence was found for the application of solid manure to potatoes (Table 1).

The within field distribution was studied for 40 fields with a drycore level >5% on tubers; with black scurf infection, the average relative risk for drycore damage on the same tuber was 2.23 times (SE 0.23) and with wireworm damage 2.46 times (SE 0.24) higher than for tubers without black scurf or wireworm damage, respectively. In contrast, no higher risk for drycore was observed on tubers with slug damage. Abiotic factors like pH, organic matter content and texture of soils as well as the meteorological conditions were without influence on drycore damage.

## Discussion

The results clearly show that *Rhizoctonia* infections alone were rarely responsible for drycore damage on potatoes. Even though infections of *R. solani* increased the risk



**Fig. 1** Percentage of tubers per field with drycore damage dependent of wireworm damage and black scurf on seed tubers on 2002 and 2003. 1 less than 5% of tubers with wireworm damage and less than 10% of the seed tubers with black scurf; 2 more than 5% of tubers with wireworm damage and less than 10% of the seed tubers with black scurf; 3 more than 10% of seed tubers with black scurf, less than 5% of tubers with wireworm damage; 4 more than 5% of tubers with wireworm damage and more than 10% of seed tubers with black scurf. The box plot shows the 50% interquartile range of the values as well as the median (middle line in the box). The points represent the outliers, and red = extreme outliers.  $N=73$  (1), 14 (2), 74 (3) and 16 (4). In order to have a global  $\alpha$ -level of 0.05 the Bonferroni–Holm procedure was applied (Holm 1979). Results of groups with different denominator are statistically different

**Table 1** Analysis of variance of drycore on potatoes for fields without fungicide seed treatment in all farming systems

Source of variation	DF	SQ	MQ	F ratio	Probability level
Wireworm (covariate)	1	0.4543	0.4543	49.41	<0.0001
Year	1	6.7938E-04	6.7938E-04	0.07	0.7865
Crop rotation	1	0.0650	0.0650	7.07	0.0094
Seed quality	2	0.1361	6.8041E-02	7.40	0.0011
Crop rotation × seed quality	2	9.7832E-02	4.8916E-02	5.32	0.0068
Manure application	1	1.4915E-03	1.4915E-03	0.16	0.6882
Crop rotation × manure application	1	1.9268E-02	1.9268E-02	2.10	0.1517
S	80	0.7357	9.1960E-03		
Total adjusted	89	1.7606			
Total	90				

Factors: years (2002 and 2003), rotation (grass clover ley 3 years before potatoes or not), seed quality (percent tubers with black scurf: 0%, <20%, >20%) manure application (yes/no), covariate: wireworm

for drycore, potato lots with heavy black scurf infestation on progeny tubers could be absolutely free of drycore. Thus, the infection of a plant with *R. solani* seems to be a prerequisite but not the only condition for the formation of drycore symptoms on tubers. Drycore mainly occurred when potatoes were wounded by wireworms (Table 1), whereas wounding by slugs had no impact on drycore. This suggests that wounds on tubers by wireworm or other means may facilitate the penetration of *R. solani* into the tuber and favour the formation of drycore. This is consistent with Schwinn (1961) who managed to infect mechanically wounded normal lenticels and reproduced typical drycore symptoms in greenhouse experiments. His second postulate that the formation of drycore symptoms is favoured under wet soil conditions in soils with higher clay content through naturally grown lenticel excrescences was not corroborated here as drycore damage occurred in all soil types independently from the clay and organic matter content and was not lower in the very dry and hot year 2003. Therefore, seed quality and crop rotation (proportion of grass clover leys) were the most important influential factors for drycore. Under Swiss conditions, the formation of drycore symptoms through lenticel excrescences seems to be of minor importance. The enhancement of wireworms by grass clover leys (Keiser et al., unpublished results) may explain in part the higher occurrence of drycore in the organic farming system as potatoes were much more often grown in this system after several years of grass clover leys. Grütte (1940) already observed that the occurrence of drycore was higher after ploughing of grassland. The fact that little research was done since Schwinn (1961) may indicate that drycore has mostly been of regional importance. However, drycore seems to be on the rise in some regions in Germany and Austria especially on organic farms which grow potatoes in crop rotations with grass clover leys (W. Dreyer, Oekoring Niedersachsen and A. Fuchs, Bioland Bayern, personal communication). The higher proportion of seed lots which exceeded the threshold for black scurf in the organic farming system and the lack of an effective seed treatment



could be further reasons for the severe drycore problem. The fact that seed quality had a significant influence on drycore infestation of harvested tubers (Table 1) indicates that soil infestation with *R. solani* was of minor importance in our survey, probably due to sufficiently long intervals between two potato crops. For more than 75% of the fields included in the survey, intervals between potatoes were more than 5 years and at least 3 years for 25%. Carling and Leiner (1986) in Alaska could not recover isolates of AG 3 from soil free of potato for 5 years and more.

Fungicide seed treatments in the integrated and conventional fields may also reduce the risk for drycore as they reduce *Rhizoctonia* infestations on plants and the infestation level of black scurf on progeny tubers. The influence of fungicide seed treatments, the higher seed quality and the use of insecticide seed treatments may explain why drycore damage occurred less in the integrated and conventional farming system even when potatoes were grown after grass clover leys.

## Conclusions

The results of our study support the hypothesis that injuries of wireworm on tubers in combination with *Rhizoctonia* infections of the potato plant increase drycore damage on potatoes. Highest drycore damage occurred when seed tubers infected with black scurf were planted and progeny tubers were wounded by wireworms. Drycore as a consequence of an infection through lenticel excrescences seems to be of minor importance under Swiss conditions. These results should be validated by field trials and pot experiments under controlled conditions.

The use of clean seed without black scurf, crop rotations with low percentage of grass clover leys and intervals of at least 3 years between grass clover ley and the potato crop are important measures against drycore. Seed quality has to be improved especially in the organic farming system where seed infestation with black scurf often exceeds the threshold and no efficient seed treatment against *R. solani* is known.

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